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ZONATION IN CULTURES OF FUSARIUM DISCOLOR SULPHUREUM

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(WITH PLATE 8)

The effect of light upon plants has received considerable attention of late. Exact quantitative and qualitative measurements have been made of the influence of light in causing phototropic turning of the coleoptiles of oats (see Nienburg, **1919**, Guttenberg, **1919**, Koningsberger, **1923**). The coleoptile appears to respond to a definite intensity of light: Guttenberg finds, for example, that twice as much light is required for the turning, if the coleoptile be shaded along half its longitudinal area, as is required with unshaded coleoptiles. It has long been known that the sporangiophores of *Phycomyces* and of *Pilobolus*, and the fruit bodies of various other fungi, turn toward the light in a manner comparable to the turning of oat coleoptiles. Tollenaar and Blaauw (**1921**) discuss the changing adaptations of sporangiophores of *Phycomyces nitens* to light, their sensitiveness depending upon the amount of light to which they had been exposed before a new light intensity was used. Oltmanns (**1907**) had also noted that the more accustomed the sporangiophores become to light, the higher is the light intensity required for the optimum phototropic response. Blaauw (**1919**) considers that in the cases of phototropic turning of coleoptiles or of sporangiophores in unilateral illumination, the phototropism is a manifestation of a localized growth response to the light; that the phenomenon of phototropism is a secondary phenomenon, the growth reaction being the primary one.

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Light is important to many fungi. Miss Westerdijk (**1923**) of the "Centralbureau voor Schimmelcultures" records that fungi in general require light, which may influence fruiting. Leonian (**1924**) found that, of twenty Sphaeropsidales which he grew in culture, no pycnidia were produced in darkness by two species, twelve others showed reduced reproduction in darkness, and only six were unaffected by darkness. The Agaricaceae may also require light: MacDougal (**1903**) found that *Coprinus stercorarius* produced only rudimentary fruit bodies in darkness, but produced normal pilei when light was allowed to act. Brefeld had previously noted similar cases. Buller (**1909**) found that the pilei of *Lentinus lepideus* and of *Polyporus squamosus* were formed as a response to the morphogenic stimulus of light, these fungi being quite sterile in total darkness. Buller also notes that in the rhythmic diurnal development of certain small *Coprini*, light acts as a causal stimulus; but he points out that certain fungi, such as *Psalliota campestris*, develop normally in darkness. *Pilobolus*, according to Buller (**1921**), possesses a pigmented "eye spot" which serves to direct the sporangium toward a source of light.

The formation of zones, *i.e.*, concentric rings, in cultures of fungi on petri dishes, etc., is a very commonly observed phenomenon. It has often been observed that one such ring is formed each day. Hedgcock (**1906**) noted these zones in cultures of *Cephalothecium*, *Penicillium*, *Mucor* and *Hormodendron* exposed to daylight or to blue light during the day. No such zonation was found in cultures grown continuously in the dark, or in red or orange light during the day. Stevens and Hall (**1909**) record that light causes the formation of zones in some fungi, although in others the zones appear to have no reference to illumination. Molz (**1906**) had found that diurnal changes in light conditions cause the formation of concentric zones in cultures of *Sclerotinia fructigena*, the conidial cushions being scattered or absent in darkness. Hutchinson (**1906–7**) also records the formation of concentric zones in *Penicillium* and *Oidium lactis* as a response to light. Milburn (**1904**) had found that concentric zones were produced in cultures of *Hypocrea rufa* in certain media, but were absent in other media, the medium, not light, being the agent

involved in the zone formation. Gallemaerts (**1910**) found that the zones in *Cephalothecium roseum* and other fungi were caused by the alternation of light and darkness, light appearing to hinder the formation of spores. He found that an all-day exposure to red, orange, or blue light, alternating with darkness caused zone formation, but that no zones appeared when the fungi were exposed to continuous light or continuous darkness. He thus differs from Hedgcock as to the effect of red and orange light. Allen and Jolivette (**1914**) state that *Pilobolus* is not strongly phototropic toward yellow light, and still less so toward red light. Miss Parr (**1918**) measured carefully the effect of different portions of the spectrum upon the phototropic response of *Pilobolus*, and found that presentation time decreases gradually from the red to the violet end of the spectrum, the time varying in inverse ratio to the square root of the wave frequency of the light.

EXPERIMENTAL RESULTS

The writer had had *Fusarium discolor sulphureum* in culture for several years. This fungus is a common cause of tuber rot of potatoes in Minnesota (Bisby, **1919**) and in Manitoba. It grows well and produces macroconidia abundantly (so long as the culture is a "high culture") in either light or darkness. Morris and Nutting (**1923**) record that light in general has no appreciable effect upon growth of *Fusaria* except in heightened color in cultures grown in diffuse light.

In working with *Fusarium discolor sulphureum*, at the University of Minnesota in 1918 and 1919, it was found that a moderately short exposure to light suffices to induce the formation of rings of spores. When cultures growing in a dark incubator were removed to the daylight only long enough to mark with a wax pencil the limits of growth, then replaced in the incubator, it was observed that by the following day (or even after about 7 hours) a ring of macroconidia could easily be seen. The ring was formed directly under the pencil mark. No effect was noted on the portions of the culture elsewhere than at the tips of the hyphae. If two exposures were made during one day, two rings were produced. If the cultures were kept in darkness, no rings resulted, yet many conidia were produced irregularly over the

culture. Figs. 1 and 2 show photographs of these cultures taken at Minnesota.

The writer recently made attempts to determine more definitely the amount of light required to produce these rings, since no reference to this point was found in the literature. In an endeavor to determine the amount of daylight required, attempts were first made with camera shutters, but the amount of light reaching the cultures was greatly reduced by the small size of the opening. Finally, the cultures on beef-extract agar with two percent glucose in petri dishes were placed in tight boxes with lids. Then by a little practice with an empty box in which the operation was timed with a watch and compared with the time of opening of the shutter of a camera, it was found that the lid could be opened and closed by hand with an approximation to 1, $\frac{1}{2}$, or $\frac{1}{4}$ second exposure to full daylight of the culture within. The petri dishes were exposed to the diffuse light from a west window in the afternoons. The amount of light of course varied from day to day and, furthermore, passed through a window pane and through the glass of the petri dish. Several trials demonstrated that under the conditions, an exposure of one-fourth to one-half second on a bright day, or one-half to one second on a dull day, resulted in the production of a ring sufficiently well marked to be easily noted by the naked eye (Fig. 3). A longer exposure led to the development of a more conspicuous ring.

Cultures kept in continuous darkness (Fig. 4) or continuous light of uniform intensity, *i.e.*, in a box lighted only by an ordinary electric light bulb of about 25 candle power, showed no rings, although many macroconidia were scattered irregularly over the cultures.

Cultures exposed to the diurnal alternation of daylight and darkness show daily bands of conidia (Fig. 5).

Temperature plays an important part in the phenomenon of zonation. At temperatures below about 18° C., few macroconidia are produced, and, therefore, no zonation can occur. Fig. 7 illustrates a culture exposed to alternate light and darkness at temperatures of 16–18° C. with no zones; the two zones shown at the periphery in the illustration were formed through the influence of light only after the temperature of the culture was raised to about 21° C.

At temperatures of 30° C. or above, a dense uniform growth of conidia (pseudopionnotes) is produced even in darkness (see Bisby, **1919**, p. 23) and exposure to light does not result in the formation of rings.

If a culture wrapped carefully to exclude all light is exposed to alternate high and low temperatures, such as a day in an ice box at about 10° C. followed by a day in an incubator at 25° C., followed by a return to the ice box, etc., zonation is obtained as illustrated in Fig. 6. This, however, is quite a different type of zonation from that resulting from exposure to light, the zones being made up of bands of loose hyphae with few conidia produced at the low temperature, alternating with bands of less fluffy hyphae bearing many irregularly scattered masses of macroconidia formed at high temperatures. Figs. 8 and 9 also illustrate the zonation produced in another *Fusarium* by exposure to alternating high and low temperatures.

In order to obtain more definite figures as to the amount of light required to produce a ring of conidia, use was made of an electric lamp, with a carbon filament standardized to give 25 candlepower. A number of cultures were kept either at room temperature, or were placed in an incubator at about 25° C. The cultures were fixed within boxes in pairs, and each box was wrapped in paper to keep out all light. The boxes were taken into a dark room, unwrapped, and placed at a distance of one meter from the source of light. The light from the 25 candlepower lamp was then turned on. It was found that approximately 6 minutes were required to produce a definite ring.

Since the standardized carbon lamp gave off a somewhat yellowish light, a tungsten-filament lamp, which gave a much whiter light, was tested and found also to be approximately 25 candlepower. When this tungsten lamp was used, it was found that an exposure of 2 to $2\frac{1}{4}$ minutes, at one meter distance, sufficed to produce a ring as sharp as that produced by the light of the carbon lamp for six minutes. An exposure of $2\frac{1}{4}$ minutes represented some 3375 meter candle seconds, a figure that may bear some approximation to from one-half to one second of daylight. Miss Jolivette (**1914**) found that a 16 candlepower tungsten lamp was more effective than a 32 candlepower carbon lamp in causing a reaction in *Pilobolus*.

In order to be sure that the effect was not produced by some factor connected with the handling of the cultures, *e.g.*, a slight change in temperature in removing the boxes from the incubator to the dark room for a few minutes, checks were used in which the boxes were handled in the same way, but with the light off. No zones were produced in petri dishes so treated. Other cultures were kept in continuous darkness: these showed no zonation.

A few tests have been made with other fungi, such as *Cephalothecium roseum*, *Penicillium* spp., *Verticillium alboatrum*, etc. While a similar type of zonation occurred with these fungi as a result of exposure to light, the zones were scarcely as sharp as with *Fusarium discolor sulphureum*. This *Fusarium* also appears to be more sensitive to light than the other fungi used.

It appeared to make no marked difference whether the light reached the culture from above or below, although of course there would be some absorption of light passing through the medium in cases in which the bottom of the petri dish was exposed.

GENERAL DISCUSSION

The production of a zone of conidia as a result of exposure to light is a response quite different from that of a phototropic turning of an *Avena* coleoptile or the sporangiophore of *Phycomyces* or of *Philobolus*. The zone comes about as a result of the subsequent formation of conidiophores and conidia at the point where the tips of the spreading hyphae receive the stimulus of light. In darkness, however, conidia are produced, but are scattered over the culture; in other words, *light results in a localization of the production of conidia in the Fusarium*. Although Hedgcock (**1906**) and Gallemaerts (**1910**) remark that light inhibits the formation of spores in the fungi with which they worked, Rahn (**1912**) considers that the greater production of spores by day, and of mycelium by night, accounts for the daily zones of mold colonies. My observations with *F. discolor sulphureum* indicate that fully as many conidia are produced in cultures receiving the short light stimulus as in those remaining in the darkness. It is, of course, true that the conidia are produced in the darkness during a few hours following the short

light exposure. We may perhaps consider that the light gives a brief check to the vegetative growth of the tips of the hyphae, and that the formation of conidiophores follows upon this check.

Fusarium discolor sulphureum develops upon potatoes in dark storage cellars, or in the soil, especially when a partially rotted tuber is planted. The production of conidia at the point where a ray of light strikes the young mycelium may serve as an aid in the dissemination of spores.

Considerably more illumination is required to produce a noticeable zone of conidia in this *Fusarium* than is required to produce a phototropic movement in a coleoptile of oats or in a sporangiophore of *Phycomyces*. The accurate measuring of the effect of different parts of the spectrum, and of precisely the intensity of light required to produce a definite zonation, would require facilities which I have not at the moment available. The production of zones is a response to light that is easily demonstrated, and a permanent record of the result can be kept by preserving the cultures.

I am glad to express my thanks to Dr. A. H. R. Buller for critically reading the manuscript.

SUMMARY

1. Zonation in cultures of *Fusaria* was found to occur from the influence of light or temperature.

2. An exposure of *Fusarium discolor sulphureum* to bright daylight for one-fourth to one-half second is sufficient to produce a ring of conidia which can be detected by the naked eye.

3. A six-minute exposure to the light from a 25 candlepower carbon lamp at one meter distance was required to produce a noticeable ring, but an exposure of 2 to $2\frac{1}{4}$ minutes to a tungsten-filament lamp of about the same candlepower sufficed to induce the zone-formation.

4. The light acts upon the outermost tips of the hyphae, and the phenomenon is, at least in part, another case of the effect of light upon growth.

5. Zonation may also be induced by alternating temperature in constant darkness.

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EXPLANATION OF PLATE 8

Figs. 3, 4, 5, 6 and 7 are from prints made directly by contact with the petri dishes containing the culture. Figs. 1, 2, 8 and 9 are from photographs made at the University of Minnesota.

Fig. 1. *Fusarium discolor sulphureum*, grown in the dark, but exposed to the light and marked before the fungus had covered the plate, with the production of a ring of conidia at the point where the tips of the hyphae were exposed. Above, a little of the mark not erased, covering the ring perfectly.

Fig. 2. A culture kept in the dark in an incubator, except when exposed a moment each day to mark the limits of growth. Some of the marks have been erased. In one case a double ring is formed as a result of two exposures, about five hours apart, in one day.

Fig. 3. Inoculated May 13, 1924. Cultures exposed to daylight by opening the box containing them, before a west window at 2 p. m. each day, as noted below. The cultures were not exposed to sunlight. The fractions of seconds are approximate.

May 15, 5 sec. bright day. Ring produced.
May 16, $\frac{1}{4}$ sec. bright day. Faint trace of a ring.
May 17, 5 sec. bright day. Well marked ring.
May 18, 1 sec. bright day. Well marked ring.
May 19, 2 sec. bright day. Well marked ring.
May 20, $\frac{1}{2}$ sec. bright day. Slight ring.
May 21, $\frac{1}{4}$ sec. dull day. No ring.
May 21, removed to full daylight at 5 p. m. Ring produced.

Fig. 4. Inoculated Mar. 31, 1924, kept in the dark continuously until the culture was grown over. No zonation produced. The radii are caused by shrinkage of the medium. Print made June 5.

Fig. 5. Inoculated May 13, 1924. Exposed to alternate day and night in the laboratory. Broad rings mark each day's growth.

Fig. 6. Inoculated July 3, 1924. Kept constantly in the dark. Alternated between an ice box at about 10° C., in which lighter hyphal areas were produced, and an incubator at 25° C., in which darker conidia-bearing areas were formed. The culture was alternated each 24 hours. (Since the print was made directly, the colors are reversed.)

Fig. 7. See explanation in text.

Fig. 8. *Fusarium ?culmorum*. Inoculated Mar. 2, 1919. Kept at about 15° C., for four days; on Mar. 6 the culture was placed out of doors, with the temperature close to freezing, and a bright red zone was produced by mycelium close to or sunken in the medium. Then placed in an incubator at 27°; then in an ice box at 10°, and finally back in the incubator. This fungus produced more mycelium at higher temperatures than at low temperatures, whereas *F. discolor sulphureum* produces spores abundantly at higher temperatures, and mycelium more conspicuously at lower temperatures.

Fig. 9. Reverse (lower) surface of the culture shown from above in Fig. 8.

BOTRYOSPHAERIA AND PHYSALOSPORA IN THE EASTERN UNITED STATES

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(With Plate 9)

Introduction

So similar are *Botryosphaeria Ribis* and *Physalospora malorum*, both in the morphology of their perfect stages and in their growth on certain culture media, that they might easily have been confused even if they occurred on but few hosts. Continued collecting and study make it apparent that they are found on a great variety of host species scattered over at least the eastern part of the United States. Moreover, both fungi may occur not only on the same host species but on the same individual or part. Stromata of the two species have indeed been found several times within the space of a square centimeter of host bark.

In an earlier paper (**5**) the writers have compared the species on apple and currant. The present paper is essentially a progress report summarizing the results of collecting and culture work to date. Although our collection of course contains a large number of specimens which show only pycnospores, attention will be confined in this paper to ascospore material in which the ascospores have been measured, the type of ascospore germination observed, pycnospores produced in pure culture from single ascospores, and the pycnospores which were produced in culture also measured. This includes seventeen hosts in the case of *Botryosphaeria Ribis* and twenty-two in the case of *Physalospora malorum*.

Botryosphaeria Ribis has been collected on the following hosts: *Amygdalus* sp. at Madison, Fla.; *Baccharis* sp., 2 specimens, both at Flamingo, Fla.; *Eucalyptus* sp. at Kissimmee, Fla.; *Gossypium* sp. at Sparta, Ga.; *Juglans* sp. at Seton, Del.; *Laguncularia* sp. at Flamingo, Fla.; *Liquidambar* sp., 2 specimens, one at Kissimmee, Fla., and one at Macon, Ga.; *Liriodendron* sp. at

Andersonville, Ga.; *Melia* sp., 3 specimens, one at Lake City, Fla., one at Monticello, Fla., and one at Fort Mott, S. C.; *Poinciana* sp. at Homestead, Fla.; *Pyrus Malus* at Falls Church, Va.; *Quercus* sp. at Lake City, Fla.; *Ribes* sp. at N. Rochester, Mass.; *Ricinus* sp., two specimens, both at St. Lucie, Fla.; *Rosa* sp. at Bell, Md.; *Sambucus* sp. at Titusville, Fla.; and *Vitis* sp. at Sebastian, Fla.

Physalospora malorum has been collected on the following hosts: *Acer* sp., five specimens, one at Orlando, Fla., one at Southern Pines, N. C., and the other three at St. Stephen, Charleston, and Aiken, S. C.; *Alnus* sp., two specimens, one at Society Hill, S. C., and one at Aiken, S. C.; *Amygdalus* sp. at Apex, N. C.; *Cercis* sp. at Madison, Fla.; *Crataegus* sp. at Aiken, S. C.; *Diospyros* sp. at Rochelle, Ga.; *Hicoria* sp., four specimens, one each at Aiken, S. C., Monks Corners, S. C., St. Lucie, Fla., and High Springs, Fla.; *Liquidambar* sp., three specimens, one at Aiken, S. C., one at Madison, Fla., and one at Thomasville, Ga.; *Liriodendron* sp. at Aiken, S. C.; *Lucuma* sp. at Society Hill, S. C.; *Magnolia* sp. at Aiken, S. C.; *Melia* sp., three specimens, one at Madison, Fla., one at Montecello, Fla., and one at Milledgeville, Ga.; *Platanus* sp. at St. Stephen, S. C.; *Prunus* sp. at Madison, Fla.; *Pyrus Malus* at Falls Church, Va.; *Quercus* sp., six specimens, one at each of the following localities: Southern Pines, N. C., Ft. Mott, S. C., Aiken, S. C., St. Cloud, Fla., Lake Alfred, Fla., and Macon, Ga.; *Ribes* sp. at N. Rochester, Mass.; *Rubus* sp. at Society Hill, S. C.; *Salix* sp., three specimens, one at Vero, Fla., one at Pampano, Fla., and one at St. Cloud, Fla.; *Sassafras* sp., two specimens, one at Ft. Mott, S. C., and one at Andersonville, Ga.; *Citrus* sp. in Baldwin Co., Alabama; *Viburnum* sp. at Aiken, S. C.; and *Vitis* sp., two specimens, one at Aiken, S. C., and one at St. Cloud, Fla.

In the paper just mentioned (**5**) the writers pointed out that the chief differences between *Botryosphaeria Ribis* and *Physalospora malorum* were the differences in the life histories, the first having *Dothiorella* as its pycnidial stage and the second *Sphaeropsis* as its pycnidial stage, and the difference in the ascospore sizes. Differences were also noted in the method of germination of the ascospores and culture characters on certain culture media.

In the present paper, evidence is presented to show that these characteristic differences are constant on the various hosts from which fresh ascospore material has recently been collected. For the present, consideration will not be given to the older ascospore material which will not now germinate.

The name *Botryosphaeria Ribis* is here used for species congeneric with *B. Ribis chromogena* of G. & D. (**2**), as represented by specimens in Fungi Columbiani *3409*. In using the name *Physalospora malorum* or *Sphaeropsis malorum* it is intended to designate the fungus causing the common black rot of apples in the eastern United States, usually and apparently correctly cited by American authors as *Sphaeropsis malorum* Peck and as represented by numerous specimens deposited by the writers in the pathological collections of the Bureau of Plant Industry and distributed in part by that herbarium. It is not intended at this time to enter into the discussion as to the specific identity or relationship of this species to *Sphaeropsis malorum* Berkeley, as found in England, or to similar species found in Europe and the northwestern United States, which have at various times been referred to as *Sphaeropsis malorum*. The writers are now inclined to believe that the safest citation for the pycnidial stage of the organism in question is *Sphaeropsis malorum* (Peck), and for the ascogenous stage, *Physalospora malorum* (Peck), instead of *Physalospora malorum* (Berk.), as previously used by them.

SPORE MEASUREMENTS

The most obvious and one of the most constant morphological differences between the perfect stages of *Physalospora malorum* and *Botryosphaeria Ribis* is the difference in the size of their ascospores. The lengths and widths of the ascospores of the specimens discussed in this and the earlier paper are summarized in tables 1 and 2. In general shape the ascospores of the two fungi are very similar indeed. It is, then, not surprising that these species have been confused, especially since the larger spores of *Botryosphaeria Ribis* equal in size the smaller ones of *Physalospora malorum* (Tables 1 and 2). The great majority of the ascospores of *Physalospora malorum* are, however, both longer and wider than those of *Botryosphaeria Ribis* and in good material

TABLE I

ASCOSPORES ARRANGED BY CLASS ACCORDING TO LENGTH

	Total number of spores	Length (microns)																											
		13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40
Botryosphaeria Ribis on *Pyrus Malus*	207	1	5	13	8	22	22	14	25	19	21	33	16	3	2	2	1												
" " " *Ribes* sp.	226	5	7	9	25	35	27	21	24	19	22	16	6	3	3	4													
" " " 15 other hosts listed in text	369	1	3	6	8	30	39	39	55	25	74	43	21	19	6														
Physalospora malorum on *Pyrus Malus*	223						1		1		5	8	4	11	10	16	11	31	20	28	24	30	11	6	3	2		1
" " " *Ribes* sp.	146								1		4	8	9	15	9	30	15	12	21	4	6	6	3	2		1		
" " " 20 other hosts listed in text	844								2		5	10	23	38	55	121	117	82	117	66	89	44	28	24	12	7	4		

TABLE II

ASCOSPORES ARRANGED BY CLASS ACCORDING TO WIDTH

	Total number of spores	Width (microns)											
		4	5	6	7	8	9	10	11	12	13	14	15	16
Botryosphaeria Ribis on *Pyrus Malus*	207	1	11	54	94	37	10							
" " " *Ribes* sp.	226		8	44	91	70	11	2						
" " " 15 other hosts listed in text	369		1	41	96	131	66	21	10	3				
Physalospora malorum on *Pyrus Malus*	223				25	31	46	68	26	23	3	1		
" " " *Ribes* sp.	146			1	2	14	23	41	33	22	9		1	
" " " 20 other hosts listed in text	844				7	30	104	222	222	162	66	26	3	2

the species may readily be distinguished by the size of their ascospores.

Not less striking than the difference in the size of the spores of the two species is the similarity in the size of the spores of each species from different hosts. While the number of spores measured from each host (except currant and apple) is not large, there is nothing to indicate that anything more than an appearance of greater accuracy would have been gained from measuring larger numbers. The agreement in the size of the ascospores from such unrelated hosts is little short of surprising, especially when it is remembered that the specimens here dealt with were collected all the way from Massachusetts and New York to southern Florida.

All the pycnospores measured were grown in pure culture from single ascospores, since with our present very limited knowledge of the species of these genera there seems to be no other method of being sure of the relation between the ascospores and the pycnospores. Merely finding them close together on a single host apparently gives no assurance that they belong even to the same genus. Pycnospores of *B. Ribis* from all seventeen hosts agree closely in size and shape, measuring 10–29 x 4–9 μ, mostly 18–20 x 5–6 μ. Pycnospores of *P. malorum* measure 17–31 x 7–15 μ, mostly 22–25 x 9–12 μ. As might be expected, agreement between pycnospores from different hosts is not as close or constant as is that of the ascospores. As yet the writers have no satisfactory information as to the extent to which the size of the pycnospores can be modified by the environment. The interesting observation of Paddock (**4**, p. 194 and 195) that the average size of spores of *Sphaeropsis malorum* varies according to the host on which they are grown should, however, be remembered in this connection.

ASCOSPORE GERMINATION

The apparently trivial character of ascospore germination has proven of great service in distinguishing the two species in culture. As noted in our earlier paper (**5**, p. 595, plate 2, figs. *O*, *P*, *Q* and *R*), under the conditions of our work in the laboratory at Washington, ascospores of *Botryosphaeria Ribis* characteristically

develop two germ tubes which branch before they have reached a length equal to ten times that of the spore, whereas ascospores of *Physalospora malorum* characteristically produce but one germ tube, which grows to a length often equal to fifty times the length of the spore before it branches. Even if an ascospore of *Botryosphaeria Ribis* develops but one germ tube it usually branches while still short. Not only has this difference remained constant throughout the course of our work on these species from all the hosts listed in this and earlier papers, but ascospores of other species of *Physalospora* with which we have thus far dealt have shown the same type of germination as *Physalospora malorum*.

While these differences have proven remarkably constant for the germination of these spores in agar plates at temperatures of 12 to 15° C., it is of course not to be expected that they would be equally constant in all media or at other temperatures. High temperatures would of course result in much more rapid germination and earlier branching. The difference noted has been very useful, however, in assisting in distinguishing these species, especially during the early part of our work.

CULTURAL CHARACTERS

In making the life history studies summarized in this paper and in their earlier work the writers have made over five thousand cultures on various media. As already noted (**5**, p. 597) cultures of these two species are much alike on some media, especially while young. They may be readily distinguished, however, on beef agar or on corn meal in flasks. Thus far the writers have failed to note any differences between these fungi as found on different hosts which would suggest that there might be actually different strains on the different hosts. The currant cane blight fungus, *Botryosphaeria Ribis chromogena*, which is chromogenic on starchy media, is the only strain yet distinguished by cultural characters, and this of course is not confined to currant.

STROMATIC CHARACTERS

Both perithecia and pycnidia of *Botryosphaeria Ribis* usually occur in a stroma. That the size of the stromata varies with the

host and the thickness of the bark was pointed out by Stevens and Jenkins (**7**) and the present writers (**5**). Perithecia of *Physalospora malorum* are usually scattered, though occasionally several are joined together. As in the case of *Botryosphaeria Ribis*, however, the size of the sporocarp of *Physalospora malorum* (**6**) is modified by the character of the host bark. On currant and hosts with similar bark several perithecia or pycnidia are grouped together in a single sporocarp surrounded by a mass of stromatic tissue, whereas on hosts with thinner bark or more uniform texture such as apple or willow, perithecia usually occur singly, and pycnidia either singly or in very small stromata.

With such modifications in both species on different hosts it is not surprising that the superficial appearance of the sporocarps of the two fungi is often so similar as to make them readily confused. When, however, mature perithecia of the two species are compared on bark of about the same thickness on the same host species, certain characteristic differences are apparent. The photographs in plate 9 give some idea of the appearance of the two fungi on various hosts when examined with a hand lens after the tops of the perithecia have been cut off.

The contents of mature perithecia in both species is almost snow white, a characteristic which makes it possible to identify the group with a fair degree of certainty in the field. As will be noted from plate 9, perithecia of *Botryosphaeria Ribis* are constantly smaller (averaging 165 by 130 μ) than those of *Physalospora malorum* (which average 245 by 210 μ) and are generally grouped in stromata. The perithecia of *Physalospora malorum* on the other hand are larger and even when they are very close together, are usually without any definite stroma. The general structure of the mature or nearly mature sporocarps suggests that interesting and significant differences would be revealed by a careful study of their development. Since, however, the two fungi occur so generally on the same host, a significant study of the development of their ascogenous stages can be made only after our technique has developed to the point where we are able to produce perithecia in quantity in pure culture.

HOST RELATIONS

The hosts on which *Botryosphaeria Ribis* and *Physalospora malorum* have already been found are sufficient in number and so unrelated as to make it seem probable that either of these species or both of them may be found on almost any deciduous host in the eastern United States. It is perhaps somewhat at variance with general practice in mycology to consider as a single species fungi found on unrelated hosts. The writers feel, however, that the practice of describing, or naming, as "different" fungi having practically identical morphological characters merely because they happen to be found on unrelated hosts is most unfortunate, particularly in the case of saprophytic or nearly saprophytic species.

In the present case there can be no justification for such action. It is indeed made impossible by the host relations of the currant cane blight fungus, *Botryosphaeria Ribis chromogena*. *B. Ribis chromogena* is readily distinguished by its striking cultural characters and by its parasitism on currant canes. Stevens and Jenkins (7) have proven by culture studies and cross inoculations that this fungus occurs on both rose and horsechestnut as well as on currant. To their work must now be added the interesting and valuable discovery of Miss Fenner (1) that *Botryosphaeria Ribis chromogena* (as well as the non-parasitic form) occurs in this country as a fruit rot of apples. If, then, this very definite and constant parasitic form occurs on hosts so unrelated as currant, horse-chestnut, and apple, it seems only natural to find that the saprophytic form also has a wide host range.

With *Physalospora malorum*, the writers have made few inoculation experiments, nor does it appear that much could be gained by so doing. Paddock (4) proved twenty-five years ago that *Sphaeropsis malorum* from persimmon (*Diopyros virginiana* L.), sumach (*Rhus typhina* L.), bitter-sweet (*Celastrus scandens* L.), choke-cherry (*Prunus virginiana* L.), hop hornbean (*Ostrya virginica* Willd.), mulberry (*Morus alba* L.), European plum (*Prunus domestica* L.), and elder (*Sambucus canadensis* L.) would produce a characteristic rot of apples and while this may by no means be taken as proving that the fungi are identical it at least suggests that host differences alone can have little

value in dealing with this species, either as regards its taxonomy or its control.

The work of Hesler (3) largely confirms that of Paddock, and proves that cultures of *Sphaeropsis malorum* from such hosts as mulberry (*Morus alba*), maple (*Acer spicatum*), and elm (*Ulmus americana*) are able to infect apple bark in the manner characteristic of *Sphaeropsis malorum* from apple itself. Hesler also calls attention to the rather remarkable fact that even as late as 1908 and 1909 it was seriously questioned whether the forms occurring on the bark, fruit, and leaves of apple were indeed one species.

The only course consistent with the information now available seems to be to refer to a single species, the material here called *Physalospora malorum* and other material having similar morphological characters, regardless of the host on which it is found. This does not mean of course that but a single species of *Physalospora* occurs on any particular host in this region. There are certainly two and probably several more, but they are not, so far as the writers have thus far observed, limited to a single host or group of hosts.

SUMMARY

Botryosphaeria Ribis is here reported from seventeen host species and *Physalospora malorum* from twenty-two host species in the eastern United States.

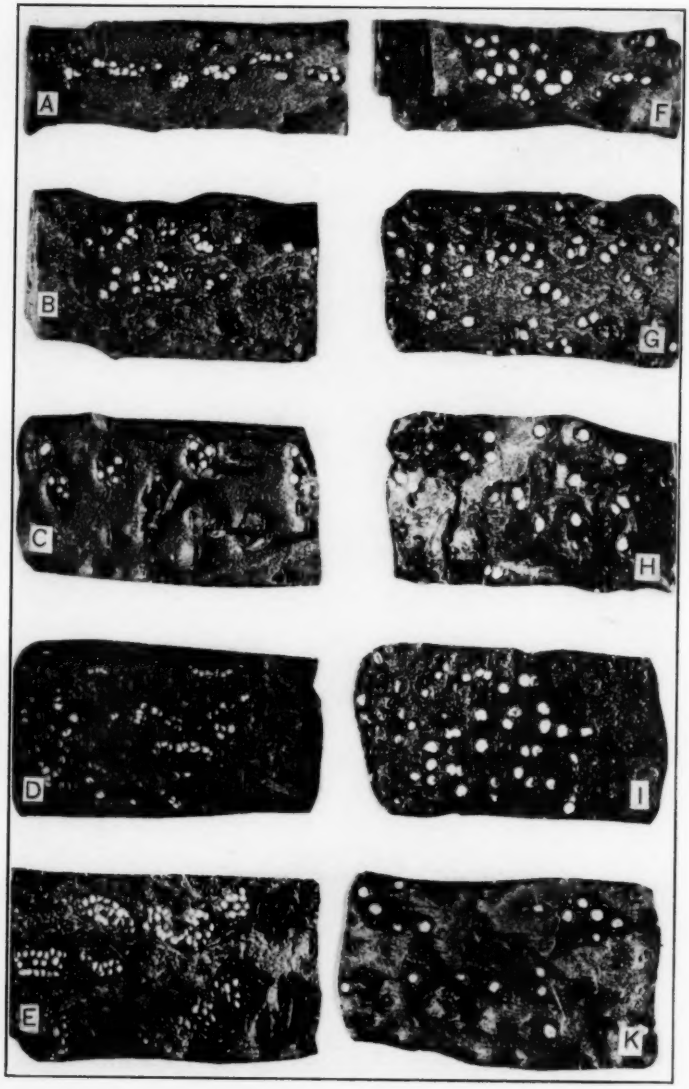
Pycnospores have been produced in pure cultures from single ascospores from each of these hosts.

Throughout this host range both species show remarkable uniformity in the size of their ascospores, the type of germination of the ascospores and in cultural characters on different media. Pycnospores, whether produced in culture or on the host, also show great uniformity.

The size of the sporocarps in both species varies with the thickness and character of the host bark.

The distinguishing characters of the two species, already pointed out in connection with our studies of these fungi on currant and apple, remain constant for all these hosts.

Ascospores of *Botryosphaeria Ribis* are constantly smaller than those of *Physalospora malorum*. Ascospores of *Botryo-*



BOTRYOSPHAERIA AND PHYSALOSPORA

sphaeria Ribis on germinating develop a short branched germ tube while those of *Physalospora malorum* develop a long unbranched germ tube. The pycnidial stage of *Botryosphaeria* is a *Dothiorella* while the pycnidial stage of *Physalospora* is a *Sphaeropsis*. Perithecia and pycnidia of *Botryosphaeria* generally occur in a stroma, while those of *Physalospora* have little, if any, surrounding stromatic tissues.

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EXPLANATION OF PLATE 9

Cut perithecia of *Botryosphaeria Ribis* on (*A*) *Liriodendron*, (*B*) *Quercus*, (*C*) *Amygdalus*, (*D*) *Salix*, and (*E*) *Melia*. × 8.

Cut perithecia of *Physalospora malorum* on (*F*) *Liriodendron*, (*G*) *Quercus*, (*H*) *Amygdalus*, (*I*) *Salix*, and (*K*) *Melia*. × 8.

MYCOLOGICAL NOTES FOR 1923 [1]

L. O. Overholts

(With Plates 10, 11)

1. Merulius rugulosus Berk. & Curt.

Dr. Burt lists this species as occurring in Cuba and Jamaica. In January, 1923, an abundant collection was made at Miami, Fla., by Mr. L. W. Nuttall. The determination was made by Dr. Burt. This is an elegant species, though more likely to be referred to *Phlebia* than to *Merulius*, since both in coloration and in the character of the hymenium it might be mistaken for a bright-colored form of *Phlebia radiata*. The unevenness of the hymenium is not in radiating ridges even in the youngest specimens, as is characteristic of that species, yet that character is likely to be obscured where several small fructifications are confluent. The color of the hymenium is a beautiful ochraceous salmon (Ridgway).

In sections (Plate 10, fig. 4) the gleocystidia are the characteristic structures, filled as they are with a golden oily substance. In many parts of the fructification there are many large imbedded crystals on the upper side of the subhymenial region.

2. Polyporus fruticum Murrill.

This species, reported by Murrill as found in Cuba, was collected by Mr. L. W. Nuttall at Miami, Fla.

3. Poria ambigua Bres.

An extended description of this species was given in one of my recent papers. Specimens have since been received from Meadville, Pa., from Taylor's Mills, Ga., from Fargo, N. D.,

[1] These notes on new and little known fungi are a continuation of similar short papers begun in 1919 and published for that year in Mycologia **12**: 135–142. 1920; for the year 1920 in Bull. Torrey Club **49**: 163–173. 1922; and for the years 1921–22 in Mycologia **16**: 1924.

Contribution from the Department of Botany, The Pennsylvania State College, No. 47.

and from Corvallis, Oregon. These localities each add a new state and extend the known range of this species very considerably. One of these collections was taken from *Ulmus*—an extension of the host range also.

4. CINTRACTIA JUNCI (Schw.) Trel.

This species was collected in abundance at Tolland, Colo., alt. 9000 ft., July 17, 1923, by C. M. Roberts, on a species of *Juncus*. I had not found it in my previous collecting in that state and it is not contained in Dr. Kauffman's recent extensive list.[2]

5. DICTYOPHORA RAVENELII (Berk. & Curt.) Burt.

Over a period of several years there have been noted in the vicinity of State College, in situations favorable to the growth of this species, particularly in old sawdust piles, small tuber-like sclerotia of a delicate pink color and connected by conspicuous strands of mycelium. When these were first found they were sectioned in order to obtain a clue to their identity but they were found to be solid masses of parenchymatous or pseudo-parenchymatous tissue. These bodies were sometimes found in the spring of the year and sometimes in early autumn, but it was not until October, 1923, that Mr. C. M. Roberts brought in specimens (Plate 11, fig. 5–6) that showed unmistakably the organic connection between these sclerotia and the fruiting bodies of *D. Ravenelii*.

In size these sclerotia vary from those (probably just beginning to develop) scarcely more than 1 mm. in diameter to others of a maximum diameter of about 1 cm. and a maximum length of about 3 cm. As shown in the illustration, these larger and more elongated forms are characteristically constricted and lobed, and show the definite connection between the sclerotia and the developing eggs. In fact the small egg in fig. 6 has a slight suggestion of having developed directly from one of these sclerotia, though such is probably not the case. Some of the more mature eggs were kept in moist chambers until they expanded into the characteristic fructifications of *D. Ravenelii*, though this fact had already been surmised from finding such fructifications

[2] Kauffman, C. H. The mycological flora of the higher Rockies of Colorado. (Papers Mich. Acad. Sci. **1**: 101–150. 1921.)

developed along with the sclerotia in the field. The consistency of these sclerotia is about that of similar tuberous bodies developed by other plants, being rather watery and somewhat fleshy. On drying, however, they become as hard and horny as do the eggs of this species when they are dried. I have not found in the literature of this species previous reference to these bodies.

6. HELVELLA INFULA Schaeff.

The half dozen previous collections that I had made of this species were made in the months of May and June, and always on the ground. At Charter Oak, Pa., on October 5, 1923, a *Helvella* (Plate 10, fig. 1) was found growing on a water-soaked, though not badly rotted, log of hemlock. The specimen was sent to Dr. Seaver who pronounced it *H. infula*. I find no previous record of its occurrence so late in the year or on such a substratum, though in exceptional years some related spring species are likely to reappear in the autumn.

7. SPHAERONEMA ACERINUM Peck.

Pycnidia columnar, elongate, or finger-like, erect, 1–2 mm. high, $\frac{1}{3}$ to $\frac{1}{4}$ mm. diameter though narrowed to a point above, the base buried in the outer layers of the woody cortex, erumpent, externally white pruinose or granular-incrusted, weathering to sordid or almost black, drying hard and brittle; ostiole not apparent; internally with a large elongated cavity extending well to the base of the column, lined with conidiophores, the wall composed of thickly arranged, vertical, branched hyphae, of which the outer layers are brown in color and more loosely arranged and studded externally more or less with calcareous-appearing matter; conidiophores simple, about 30 μ long, 2–3 μ diameter; spores oblong or short cylindric, hyaline, one-celled, 17–20 x 5–6 μ.

Bursting through the bark of dead poles of *Acer rubrum*. Stone Valley, Huntingdon Co., Pa., Sept. 30, 1923. *Overholts 8846*. (Plate 10, fig. 2–3.)

This species was originally described by Peck from three localities in New York state My collection was determined by Professor Dearness.

8. **Mycosphaerella smilacicola** (Cooke) comb. nov.

Perithecia mostly epiphyllous, a few hypophyllous, black, rather abundant, globose or subglobose, 75–105 μ diameter, imbedded in the mesophyll of the leaf, finally barely rupturing the epidermis; perithecial wall black, cellular, 8–12 μ diameter, 3 to 4 cells thick; asci clavate-cylindric, mostly with acutely pointed tips, 30–40 x 8–10 μ; spores 8 per ascus, mostly obliquely uniseriate, clavate or cylindric-clavate, two-celled, one cell usually narrower than the other, hyaline, 15–18 x 3.5–4 μ; paraphyses absent.

On leaves of *Smilax;* occupying definite rounded dead spots, 5–12 mm. in diameter, the spots brown in color, with a raised purplish or deep red-brown marginal band 0.5–1 mm. wide. Taylor's Mills, Ga., May 19, 1922. J. C. Dunegan *612. Overholts 9203.* (Plate 11, fig. 7, 8.)

The history of this species is slightly confusing. Schweinitz [3] originally described it as *Sphaeria smilacicola* and reported it as on the leaves of *Smilax rotundifolia* in Pennsylvania and South Carolina. However, he had previously described [4] another fungus under the same name as occurring on *Smilax* stems. This latter fungus is now referred to *Diatrype* and is entitled to the specific name *smilacicola.* Cooke [5] later described what is apparently the present species as *Sphaerella smilacicola. Sphaeria smilacicola,* therefore, as applied to the leaf fungus becomes a homonym, but fortunately Cooke's description of the same species as *Sphaerella smilacicola* allows the specific name to be retained unless it is antedated by some other. For the old generic name *Sphaerella, Mycosphaerella* would now be substituted. Apparently Cooke had before him numbers *95* and *155* of Ravenel's Fungi Americani when he studied this species. Yet these can hardly be considered as authentic specimens of Schweinitz's species. Ellis and Everhart [6] report that the types of Schweinitz as preserved at Philadelphia are sterile with no perithecia developed. This is an additional reason for basing the species on Cooke's description in Grevillea.

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[3] Trans. Am. Phil. Soc. **4**: 226. 1834.
[4] Trans. Am. Phil. Soc. **4**: 196. 1834.
[5] Grevillea **6**: 129, 146. 1878.
[6] North American Pyrenomycetes, 747. 1890.

EXPLANATION OF PLATES

PLATE 10

Fig. 1. Photo, × 5/7, of ascocarp of *Helvella infula*. *Overholts 8774*.

Fig. 2. Low power microphoto of free-hand vertical section through the columnar pycnidium of *Sphaeronema acerinum*, showing the internal cavity continuous to near the base of the column. The tip of the column has become recurved in sectioning. *Overholts 8846*.

Fig. 3. High power microphoto of a small portion of the same section shown in Fig. 2, showing the construction of the pycnidial wall, lined internally with conidiophores producing the oblong conidia.

Fig. 4. Microphoto of a free-hand vertical section through the fructification of *Merulius rugulosus*. *Overholts 8682*.

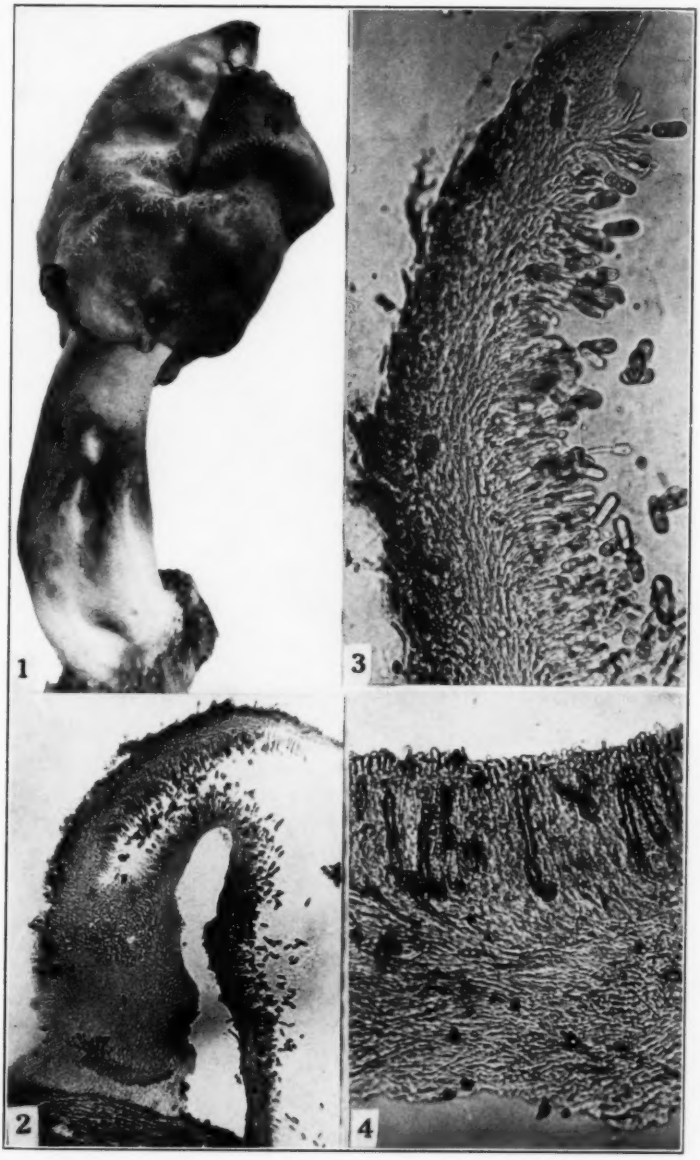
PLATE 11

Fig. 5. Photo, × 1, of a sclerotium and an egg of *Dictyophora Ravenelii*, with a mycelial strand connecting them. *Overholts 8793*.

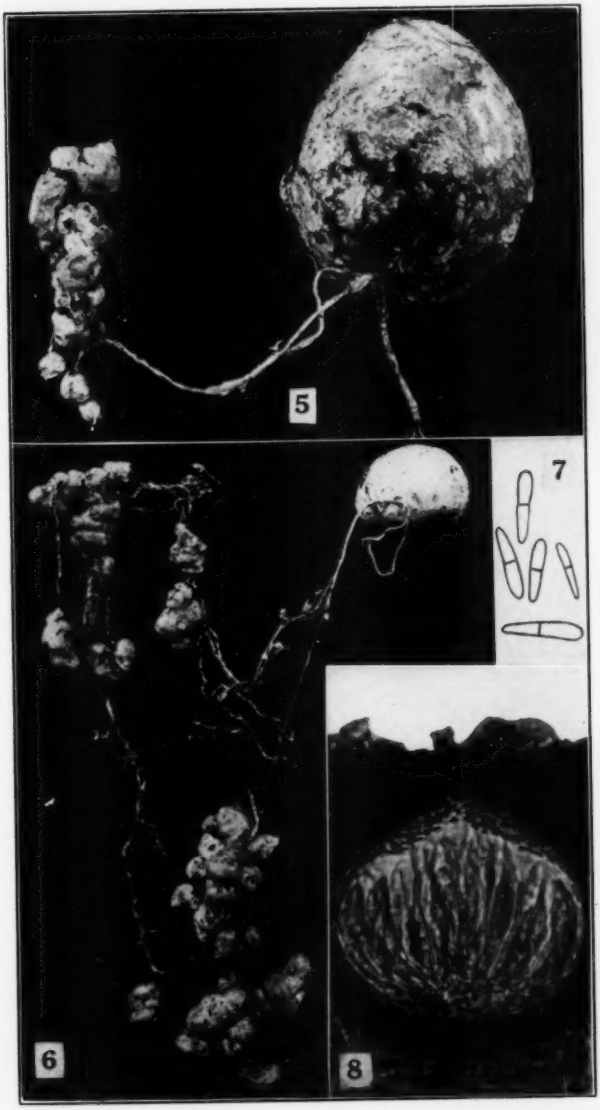
Fig. 6. Photo, × 1, of several sclerotia of *Dictyophora Ravenelii* connected by mycelial strands. Also in the upper right hand corner a small egg of the same species.

Fig. 7. Ascospores of *Mycosphaerella smilacicola*.

Fig. 8. Microphoto of a free-hand section through the perithecium of *Mycosphaerella smilacicola*. *Overholts 9203*.



1. HELVELLA INFULA. 2–3. SPHAERONEMA ACERINUM. 4. MERULIUS RUGULOSUS



5–6. DICTYOPHORA RAVENELII. 7–8. MYCOSPHAERELLA SMILACICOLA

THE GENUS GOMPHIDIUS IN THE UNITED STATES [1]

C. H. KAUFFMAN

(WITH PLATES 12–14)

I reviewed the salient characteristics of the genus *Gomphidius* in the introduction to the genus in the Agaricaceae of Michigan, 1: 170 (**8**). At that time only a few species had come to my notice in the field in this country, although I had seen *G. viscidus* Fr. and *G. glutinosus* Fr. growing in abundance in northern Europe. In European countries only four species of this genus are usually recognized in floras: *G. viscidus* Fr., *G. glutinosus* Fr., *G. roseus* Fr. and *G. maculatus* Fr. The British mycologists, however, report, in addition to these species, also *G. gracilis* Berk., and Rea (**12**) says the latter is "common" in fir woods and heaths. Recent accounts of the American species of this genus have been published by Murrill (**9** and **10**).

The species of this genus usually possess a viscid or glutinous, and hyaline, universal veil; in some species, however, this veil is dry or merely moist, developing a slight viscidity only in very wet weather. The glutinous veil surrounds the young button, in the same manner as in the subgenus Limacium of the genus *Hygrophorus;* in the subgenus Myxacium of the genus *Cortinarius;* and in certain other genera of the Agaricaceae. In a few species, this veil is scarcely or not at all evident, although it is possible that even in these a blematogen is present in the early stages. In most species this veil forms a definite teleoblema as is shown by the fact that it is separable from the pileus as a distinct, thick pellicle. Confusion may easily arise concerning the color of the universal veil, because of its tendency to become black in a number of the species. Berkeley says (**2**) the pileus of *G. gracilis* Berk. "is covered with a smoke-colored gluten." However, the veil of this species, like that of a number of others,

[1] Papers from the Department of Botany of the University of Michigan, No. 221.

doubtless blackens easily when touched or in age, and in the fresh condition it is likely that the gluten is hyaline. Dr. Peck kept this distinction in mind when he described *G. nigricans* Peck (11) with its sensitive gluten. After giving the color of the pileus as "pale brownish-red," and that of the stem as "whitish," he says: "The species is usually known by the blackening gluten which smears both pileus and stem. . . . In the dried state the whole plant is black." The degree of blackening of the surface of the pileus and stem is much used in the descriptions of the species of this genus, but it is often difficult in an examination of dried plants to apply statements concerning it, because of the extreme blackening that may occur due to overheating. On the other hand, this character is of great help in discriminating between certain species when they are properly dried. Since the blackening of cap and stem is due to the veil, it is at once evident that rain-washed plants may not show this reaction after the specimens have been dried. Certain species, however, when in growing condition, do not dry black when properly dried. Such are *G. tomentosus*, *G. furcatus*, *G. subroseus* and *G. viscidus* forma *columbiana;* others like *G. ochraceus*, *G. maculatus* and *G. glutinosus* show this characteristic in greater or less degree, while *G. nigricans* shows it to an extreme extent.

A cortina-like partial veil, more or less continuous on the inside of the universal veil where the latter passes over the gill area, is present in most and perhaps in all species in the young condition; it is never copious and is soon evanescent.

The *gills* are decurrent, subdistant or distant; in only one species, viz., *G. vinicolor* Peck, can the gills be said to be close. The type of this species at Albany clearly has closer gills than is usual, but whether this character is consistent in the fresh plant is not too certain. Poor, small or undeveloped specimens of the other species sometimes give the effect of "close" gills.

The color of the gills changes markedly during the development of the spores. If one could make a color chart of these changes for each species, I am convinced that the shades of color represented would be quite specific for each species. I have taken down these shades of color at the time of collecting the fruit-bodies and found that even in the same region and with the

same undoubted species, I would get such a variety of Ridgway color names as to be more confusing than helpful. This is due to the fact that one necessarily collects the plants at different stages of maturity. Giving a general color description seems to be the only useful procedure, but even thus, the specific differences can be brought out. It is desirable to obtain the color of the gills before the fruit-body is overmature, because the very mature gills of the different species may be much alike. For example, the gills of *G. furcatus* Peck are "testaceous" (Ridg.) at a certain stage, and those of the new form, *G. viscidus* described in this paper, are "tawny" and later "sepia" in color. The European species *G. glutinosus* and *G. viscidus* should be distinguishable by their gill-color according to the emphasis (in italics) usually placed on the colors given, but unfortunately there is no exact agreement among European mycologists; and as explained above, this is not surprising when not all the stages are given. Reading between the lines, it seems evident that the prevailing color of the gills of *G. glutinosus* is some shade of gray, while those of *G. viscidus* tend to have tawny shades which become clouded with olivaceous or purplish hues.

The gill-trama is said by Fayod (**4**) to be divergent ("bilaterale"). My own records are not complete for all of the species, but in my collection of *G. glutinosus* in Idaho, the gill-trama was composed of interwoven floccose hyphae of large cells. The texture of the gill-trama is quite mucilaginous and tends to disintegrate.

The spores of all the species of *Gomphidius* known are elongated, fusiform or subfusiform to subcylindrical, and generally large in size. The largest spores were reported by Dr. Peck for his species *G. flavipes;* the maximum length recorded by him was 30 μ. In examining the type, which was dried too hard and is not very satisfactory, I found enough spores measuring 25 μ in length to make it very probable that the extremes are 30 μ. The color of the epispore as seen under the microscope can best be called sooty varying in intensity in different species or during different stages in the maturity of the spores. In some species the epispore appears punctate under high magnification.

In Fayod's account (**4**) of the generic characteristics of *Gomphidius*, due attention was given to the *cystidia*, which other authors since then have all too often neglected. These remarkable structures, cylindrical, large, and protruding markedly above the hymenium, Fayod considered as a unique means of distinction, separating this genus from all the others. As seen from the synopsis following, he was nearly correct; only *G. oregonensis* Peck and *G. nigricans* Peck have so far been examined by me in which the hymenium either lacks these characteristic cystidia or relatively few occur. Atkinson (**1**), in his account of *G. nigricans*, does not mention any cystidia although I was able to locate a few in my own examination of his material. There is little doubt that Atkinson had the true *G. nigricans*. In all my mounts of Peck's type material, I was unable to locate any cystidia.

Fayod, in his description of the cystidia (l.c.), mentions a crust-like covering over the main body of the cystidium, and illustrates this characteristic in his figure of the cystidia of *G. viscidus* (Pl. 6, fig. 7*b*). In the figure referred to, this crust appears to be of a crystalline nature. Neither Ricken (**13**) nor Rea (**12**) mention this peculiarity of the cystidia in the European species, although the cystidia are described by these authors. In all the material which I have examined, which included most of the species in the fresh condition as well as all of the American type collections, I have rarely seen anything approaching Fayod's figure. The cystidia are extremely abundant in some species and hence such a character should be easily seen. However, occasionally one does find a sort of sheath, of a somewhat roughened or crystalline character, but normally the wall is entirely smooth. Therefore I am inclined to consider this encrusted condition a fleeting or developmental condition, not to be used as a specific character.

The relationship of the genus *Gomphidius* is not very clear. In the opinion of Fries (Monographia **1**: 149) "they have the stature of Limacium, but seem to hold closest relationship with *Cortinarius*, from which, nevertheless, they are far removed by the nature and color of their spores" (**7**). In Hymenomycetes Europaei (**6**), p. 399, he placed the group between *Cortinarius* and *Paxillus*.

Fayod (**4**) argues for a relationship between *Gomphidius* and *Paxillus* and includes both these genera in his tribe Paxillés. He, as well as others, laid a foundation for his argument by keeping Schweinitz's American species "*Agaricus rhodoxanthus*" (**14**) in the genus *Gomphidius*. This plant has been fully discussed by Atkinson (**1**) who places it in *Paxillus*, where I have hitherto kept it. In passing, it may be well to point out that it has a much closer relationship to some of the American species of *Boletinus*, and might well be put into that genus, disregarding its gill characters (the gills sometimes anastomose), and taking into account its habit, texture, spores and cystidia as of more relationship value than the gills. Such a situation has arisen with regard to the species of *Lenzites*, and a number of mycologists now agree to attach the latter genus to the Polyporaceae, a procedure which seems to me thoroughly scientific. Fayod (l.c.) pointed out the similarities of the Schweinitz plant with species of *Boletus*, especially with *B. subtomentosus* Fr., but as far as I know no one except Battaille (Les Bolets, p. 24, 1908) has definitely placed it in that group. The plant should be called *Phylloporus rhodoxanthus* (Schw.) Bres. (**3**), and placed next to the genus *Boletinus* in the Boletaceae.

Ricken has grouped *Gomphidius* with *Hygrophorus*, indicating its relationship with the subgenus Limacium of that genus, a disposition which I have followed (**8**), and which seems to me to have much in its favor. The structure of the veils, pileus, stem and gills are practically alike in certain species of *Hygrophorus* and of *Gomphidius*, the outstanding difficulties being the necessity of accounting for the characteristic cystidia and spores of the latter. But I can see no other connection where the difficulties are not more numerous. Rea (**12**) has placed it next to and presumably "above" the genus *Flammula*, but this arrangement seems to me entirely unsupportable.

Our species are found practically always in the neighborhood of coniferous trees, often in sphagnum bogs or in deep moss in the forest. Collections are made infrequently except in the northwestern Pacific coast states, where several species are quite abundant.

I wish to express my obligations to the authorities of the

New York Botanical Garden and to Dr. House of the New York State Museum for the privilege of access to the types of Dr. Murrill and Dr. Peck; also to the Department of Plant Pathology of Cornell University for allowing me to examine all of the specimens in the Atkinson herbarium.

SYNOPSIS OF THE SPECIES OF GOMPHIDIUS OCCURRING IN THE NORTH TEMPERATE REGIONS

1. Cystidia few or lacking; plants becoming black when dried. (*G. glutinosus* may be sought here) 2
1. Cystidia present and more or less abundant, long and cylindrical 3
2. Spores 15–22 x 5.5–6.5 μ; pileus 2–7 cm. broad, not umbonate, glutinous, pale brownish-red when fresh; eastern U. S. (See figs. 50 and 51, Atkinson 1) ..... *G. nigricans.*
2. Spores 10–13 x 3.5–4.5 μ; pileus 5–10 cm. broad, not umbonate, viscid, livid flesh-colored when fresh, becoming black-spotted; stem citron-yellow below ..... *G. oregonensis.*
3. Pileus with ochraceous shades, at least when young and fresh; western U. S. 4
3. Pileus not noticeably of these colors 5
4. Pileus glabrous and glutinous; spores 15–19 (22) x 6–7 μ. (See description) ..... *G. ochraceus.*
4. Pileus densely floccose-tomentose, dry or nearly so; spores 17–21 (24) x 6–9 μ ..... *G. tomentosus.*
5. Pileus 5–10 (12) cm. broad 6
5. Pileus 2.5–5 (6) cm. broad 7
6. Pileus convex to plane, not umbonate, glutinous, livid purplish-brown; gills at first whitish, becoming gray to blackish at maturity; cystidia few to scattered; spores 17–22 x 6–7 μ ..... *G. glutinosus.*
6. Pileus with subconic umbo, only slightly viscid, dark vinaceous-red to dark reddish-brown; gills at length olivaceous to purplish-umber; cystidia abundant; spores 17–20 (21) x 6–7.5 μ ..... *G. viscidus.*
7. Stem with yellow base or yellow more or less throughout, concolorous within 8
7. Stem not yellow 12
8. Pileus 2.5–6 (7) cm. broad 9
8. Pileus 1–2.5 cm. broad, convex-plane, pink, with white flesh; gills distant; spores 22–25 (30) x 6–7.5 (8) μ ..... *G. flavipes.*
9. Gills more or less forked 10
9. Gills not forked or very rarely; stem remaining dull red and pileus sooty-red when dried; pileus with subconic umbo; spores 16–21 (23) x 6–7 (7.5) μ. (See description) ..... *G. viscidus* forma *columbiana.*
10. Pileus not umbonate; stem 6–12 mm. thick 11
10. Pileus umbonate, pale vinaceous-brown when fresh, gluten becoming smoky; stem 3–6 mm. thick; spores 16–19 x 5–7 μ (Rea) ..... *G. gracilis.*
11. Stem sheathed by the viscid veil, white upwards and remaining whitish when dried; pileus salmon-colored to vinaceous-pink; spores 14–17 (20) x 5–6 μ. (See description) ..... *G. subroseus.*

11. Stem not sheathed, at length becoming black-stained or blackish when dried; veil lacking or obsolete; pileus brownish-incarnate, at length black-stained; spores 15–23 x 6–7.5 μ. . . . . *G. maculatus.*
12. Gills distinctly forked; stem attenuate downwards or pointed at base. . 13
12. Gills not forked, close; pileus dark red; stem vinaceous-reddish, not pointed at base; spores 17–22 x 5.5–6.5 μ. (Type). . . . . *G. vinicolor.*
13. Spores 15–17 x 4–5 μ (Rea); pileus rose-colored; stem whitish, tinged rosy at base; gills whitish-cinereous, finally olivaceous. . . . . *G. roseus.*
13. Spores 16–21 (24) x 5–6.5 (7) μ; pileus at first whitish, then "testaceous" (R.); stem white, becoming pinkish; gills whitish, becoming testaceous, at length variegated with sooty-brown. . . . . *G. furcatus.*

UNDESCRIBED AND EMENDED SPECIES

Gomphidius ochraceus sp. nov.

Pileus 3–6 cm. broad, convex-expanded then plane, *glabrous, glutinous,* "ochraceous-salmon" to "apricot-orange" (Ridg.) when fresh, soon clouded with "olive-brown" and gradually becoming tinged with vinaceous shades, even or soon rugose-wrinkled from the drying gluten, margin at first incurved, at length spreading; flesh thick, abruptly thin on margin, whitish to "pinkish-buff." Gills decurrent, subdistant to distant, rather broad, 7–8 mm., "tawny" (Ridg.), thick, edge entire. Stem 8–10 cm. long, tapering downwards, 6–10 mm. thick at apex, variously curved, subviscid, solid, "orange-buff" to "zinc-orange" (Ridg.), more or less floccose upwards to an obsolete annulus, concolorous within. Spores ellipsoid-fusiform, 15–19 (22) x 6–7 (8) μ, smooth, pale smoky. Cystidia abundant, cylindrical, hyaline, rounded-obtuse at apex, 150–180 x 12–15 μ. Odor none. Taste mild. Closely gregarious.

On deep moss under conifers, near Welch's Post Office, Oregon National Forest, Mt. Hood, Oregon. September 29, 1922. Collected by C. H. Kauffman. Type in the herbarium of the University of Michigan.

This species has such a superficial similarity to *G. tomentosus* Murrill that it is easily passed by as that species. It differs definitely, however, in its glabrous and glutinous pileus, and in its somewhat broader and more distant gills. *G. tomentosus,* which was abundant in this region, and was carefully studied, does not have what I should call "distant" gills as given in Murrill's description; rather the gills are close to subdistant. The color is not very sharply different, but a difference does show whenever the fresh plants are compared. The subviscid stem indicates a thin and evanescent universal veil.

Gomphidius subroseus sp. nov. (Plate 13).

Pileus 3–6 (7) cm. broad, convex-expanded then plane, very obtuse to broadly depressed, with a viscid separable pellicle, glutinous only in rainy weather, varying "salmon-color" to "vinaceous-pink" (Ridg.), disk "ochre-red" to "testaceous," usually fading, becoming slightly blackish on drying, glabrous, even or slightly wrinkled when dry; flesh thick on disk, abruptly thin on margin, white or tinged vinaceous. Gills decurrent, close to subdistant, attenuate at ends, 6–7 (8) mm. broad at middle, soon "pale smoke-gray" to "pale mouse-gray," finally darker and variegated, some forked near margin or towards stem, thickish, edge entire. Stem 3–6 cm. long, 6–12 (15) mm. thick, tapering downwards or subequal, straight or curved, solid, base or lower half "lemon-yellow," "empire-yellow" or "citron-yellow" (Ridg.), apex or upper portion white and silky, covered when fresh up to near the apex by the hyaline, viscid, thin, appressed sheath of the universal veil, at length glabrous and dry, rarely becoming sordid or blackish in age. Spores 14–17 (20) x 5–6 μ, subfusiform-ellipsoid, obtuse at ends, smooth, dark sooty. Cystidia rather abundant, cylindrical above the slender pedicel, hyaline (in fresh plants), 100–140 x 8–15 μ, apex rounded. Odor and taste none.

Type on humus and moss under conifers, near Welch's Post Office, Oregon National Forest, Mt. Hood, Oregon. September 22, 1922. Frequent in this region. Also under pines, Tolland, Colorado, September 14, 1920; and near Copeland, Idaho, September 2, 1922. Collected by C. H. Kauffman. Deposited in the herbarium of the University of Michigan.

This species differs from *G. roseus* (Fr.) Quél. by the distinct yellow base of the stem, by the less rosy-red color of the pileus attributed to the European plant, and perhaps by the cystidia. We apparently have no account of the cystidia of *G. roseus*. Ricken, to be sure, gives cystidia for the plant he places under that name, but Ricken's description departs from the conception of other mycologists, and his plant may be the species here described, or perhaps it is *G. gracilis*. Rea (**12**) unaccountably copies Ricken's remarks on the cystidia, but Rea's description otherwise applies to the plant with a rosy stem-base, and is therefore to be considered the correct traditional conception of *G. roseus*.

G. gracilis Berk., although it has a yellow stem-base, is described by Berkeley as having a conic-hemispherical pileus and

others agree that it is more or less umbonate; *G. subroseus* has a rounded pileus from the beginning and later becomes plane to depressed. After the specimens were dried they turned somewhat blackish, but when fresh this tendency to blacken—so noticeable a characteristic in some species—is very slight in this western species. From *G. flavipes* it is readily distinguished by its smaller spores, and from *G. maculatus* by the veil on the stem.

GOMPHIDIUS OREGONENSIS Peck (emended).

Pileus 5–10 cm. broad, at first convex, obtuse, becoming plane, glutinous from the universal veil, livid flesh-colored when fresh, becoming black-spotted in age and blackish when dried, glabrous, even; margin at first incurved. Gills short-decurrent, close to subdistant, gray when partly mature, then blackish. Stem 3–8 cm. long, subequal or tapering downwards, rather stout, 8–15 (25) mm. thick, floccose-fibrillose, citron-yellow almost to the apex, sheathed in part by the glutinous veil which sometimes terminates in a glutinous ring near the apex, yellow within at the base, surface becoming black-spotted when handled. Odor and taste mild. Spores narrow, elongated-ellipsoid, 10–13 x 3.5–4.5 μ, smooth, tinged smoky. Cystidia few, subcylindrical, 100–125 x 15–18 μ.

Description drawn from fresh plants collected at Lake Cushman, Washington, 1915, by C. H. Kauffman.

This has much the habit and stature of *G. glutinosus*, but is definitely distinct by its relatively small spores, and the somewhat different colors when fresh. The glutinous veil is quite thick on the pileus and especially so on the incurved margin of the young fruit-body. It is reported from all the Pacific coast states.

This species was incompletely described by Dr. Peck, who apparently drew his description from dried plants. Few collections are in the eastern herbaria that I examined. The specimens from California, distributed under this name by C. F. Baker in "Pacific Coast Fungi, No. *155*," is not a *Gomphidius*, but probably a *Paxillus* with globose spores—at least this is true of the copy in the Atkinson herbarium. On the printed label of this number, Baker states that the gills are phosphorescent, a statement which is therefore not dependable in its application to *G. oregonensis* (**9**).

Dr. Lane of Portland, who sent the specimens from which Dr. Peck drew his description, wrote Dr. Peck that this species "grows there by the wagon load." Murrill (l.c.) says he "found it common both in Washington and Oregon." Zeller (**15**) also reports it as "one of the very common Agarics" around Corvallis, Oregon. My experience differs for the localities I visited in Washington and Oregon. At the base of Mt. Hood, *G. tomentosus* was very abundant, and rarely one could pick up also a few specimens of *G. ochraceus* and *G. subroseus*. In the Cascade range east of Seattle, only *G. tomentosus* was found. In northern Idaho the latter species also occurred. In the Olympic Mountains, however, in addition to the frequent *G. tomentosus*, I obtained two solitary-growing specimens of *G. oregonensis*. The questions then arise, is it *G. tomentosus*, instead of *G. oregonensis*, which is so common in these regions, or is each common only in certain localities? The simple process of determining the spore-size will doubtless be sufficient in the future to decide these points.

GOMPHIDIUS VISCIDUS Fr. **columbiana** form. nov.

Pileus 2–6 cm. broad, at first subconic-campanulate then expanded-umbonate, subviscid, or viscid in wet weather, color when fresh "auburn," "bay" or "Hay's russet" (Ridg.), sometimes with purplish tints, very glabrous, even, shining when dry; margin at first incurved, and cortinate with an evanescent, "apricot-buff" cortina; flesh thick on disk, abruptly thin on margin, tinged pinkish. Gills decurrent, broad in middle, narrowed towards ends, close to subdistant, distinct, none or very few forked, thick, soon "ochraceous-tawny" to "tawny," at length "sepia" or "Prout's brown" (Ridg.). Stem 3–7 (8) cm. long, 4–12 (15) mm. thick, subequal or ventricose downwards, or somewhat pointed at base, solid, varying when young from "capucine-buff," or "flesh-ochre" to "apricot-orange," at length sordid brown, concolorous within, when fresh covered with delicate, appressed fibrillose shreds, glabrescent except at the obsolete cortinate zone at apex, which is colored by the spores. Spores 16–21 (23) x 6–7 (7.5) μ, subfusiform, smooth, tinged smoky. Cystidia very abundant, cylindrical, with slender pedicel, hyaline, 120–150 x 15–18 μ. Odor and taste none.

Description from studies in the field. Collected in the Rocky mountains of Wyoming and Colorado. August and September. The American form of *G. viscidus* differs from the European plants

in the much smaller average size, somewhat different colors, and a tendency to form longer and more truly fusiform spores. Exsiccati from Europe which I examined came from Sweden, France, and the mountains of Italy; some are in the Atkinson herbarium and some at the New York Botanical Garden. When properly dried, the stems of the European form are regularly and conspicuously "ochraceous-tawny" to "cinnamon-brown" (Ridg.). The dried stems of the American form, when not darkened by overheating, are always dull reddish; the caps also become this color but tend to assume darker shades of it. Well-dried plants of the two forms do not "match." This American form occurs definitely throughout the Rocky Mountain and Pacific coast states. Specimens from California slightly larger than usual are in the herbaria mentioned. Whether any of the collections from the eastern United States usually referred to *G. viscidus* actually belong here, I am unable to say.

COMMENTS

GOMPHIDIUS FLAVIPES Peck.—This must be a rare species. The type specimens, as Murrill (Mycologia **14**: 125) has already pointed out, are of no value for comparison. The unusually long spores are its principal claim to recognition.

GOMPHIDIUS FURCATUS Peck.—Apparently a species of the eastern United States only. It is probable that collections referred at times to *G. viscidus* Fr. belong here. The spores of the type material at Albany vary slightly longer than the length given by Peck. (See synopsis in this paper.) I have found this twice in Maryland, under pines; the pileus of these was 2–7 cm. broad, the stem up to 10 cm. long by 3–10 (12) mm. thick. Peck's plants had more slender stems. The color of the fresh pileus is "testaceous" (Ridg.), of the stem "congo-pink": after drying the color of both is pale reddish. Small specimens do not always have the gills forked. This species differs from all forms of *G. viscidus* in the absence of yellow in the stem. At Albany, there is a collection by Earle from Alabama, marked "*G. alabamensis* Earle." This is very probably *G. furcatus*. It grew "among needles under pine," has forked gills and the notes say that it was "pale reddish-brown throughout."

GOMPHIDIUS GLUTINOSUS Fr. (Plate 12).—There are good specimens from Bresadola at the New York Botanical Garden, and typical plants from Sweden in the Atkinson Herbarium. The principal point to be brought out here is that the cystidia are not abundant. In fact in some mounts it is difficult to locate them. In overmature and dried plants they are frequently shriveled, so that observations must be carefully checked. The cystidia of this species also appear to be less cylindrical than in many of the others. The caps are usually quite large, and the stems stout and long. It may be said to be the largest species.

GOMPHIDIUS GRACILIS Berk.—I have a few collections from the mountains of Washington which are referable to this species. There are, however, slight differences and further notes are needed. I have seen no European specimens. Our western plants have longer and stouter although rather more slender stems than described for the European species. The spores of the western plants measure 15–18 x 5–6 μ, and the cystidia are abundant.

GOMPHIDIUS MACULATUS Fr.—I have found no facts which make it necessary to change my account of this (**8**, p. 170). The comments given with that description on other species (l.c.) are, however, revised in this paper.

GOMPHIDIUS NIGRICANS Peck.—This is an eastern species, seldom collected. (See remarks on p. 114.)

GOMPHIDIUS ROSEUS (Fr.) Quél.—(See remarks under *G. subroseus*.)

GOMPHIDIUS TOMENTOSUS Murrill (Plate 14).—(See remarks under *G. oregonensis*.) The following descriptive data may be added to Murrill's account (Mycologia **4**: 307. 1912). Pileus 3–7 cm. broad, obtuse, sometimes actuely subumbonate, the thick pellicle separable and slightly viscid in wet weather, "ochraceous-buff" (Ridg.), darker when wet and then "vinaceous-tawny" to "wood-brown" (Ridg.), deeper ochraceous when dry; flesh thick on disk, abruptly thin on margin, whitish to ochraceous, shot through with "pinkish-buff" hues. Gills close to subdistant, 6–8 mm. broad in the middle, "ochraceous-buff" to "ochraceous-salmon" (Ridg.), at length sooty-brown. Stem 6–12 cm. long, 8–15 (20) mm. thick, rather firm and rigid, con-

color, at first floccose, then lacerate-fibrillose or denuded, sometimes slightly viscid; flesh compact, concolorous, "empire-yellow" (Ridg.) towards base. Taste often tardily but slightly disagreeable. Cystidia abundant, cylindrical, with slender pedicel which extends below the hymenium, hyaline, rounded at apex, 150–180 x 10–15 (18) μ, variable in length and thickness. In dense coniferous forests of fir and hemlock.

The unique tomentose-hairy surface is due to the thick universal veil which surrounds the young unopened plant. On the stem as it elongates the veil is lacerated, broken into fibrillose shreds or washed off in some cases. Sometimes the portion of it encircling the apex of the stem persists as a floccose-hairy annulus. The inferior veil is fibrillose-silky and concolorous, soon disappearing. The base of the stem is often deeply imbedded in conifer-needle beds or in moss cushions. The spores are as given by Murrill.

GOMPHIDIUS VINICOLOR Peck.—The dried type specimens at Albany are distinctly red-brown. A collection in the Atkinson herbarium from Dr. Herbst and collected in Lehigh Co., Pennsylvania, is very probably the same, although the spores average quite a little shorter. The spores of this species are notable for their more ventricose shape and appear much more fusiform under the microscope than most, especially those of *G. viscidus* forma *columbiana* which also dries reddish-brown and of which the spores tend to be subcylindric in shape. I have spoken before (**8**, p. 171) of the tendency for small or late-growing plants of this genus to have shorter spores than in "normal" plants. Considering that Peck describes the gluten of the pileus as turning black on drying, it is surprising to find the type specimens of this species unblackened, which indicates that the gluten or viscidity is thin and disappears. The species is rare. I should hesitate to refer here the plants from around San Francisco, California, some of which I examined at the New York Botanical Garden. (See Mycologia **4**: 307.) The form mentioned by me in Agaricaceae of Michigan I, p. 171, as form "minor," is a slender little plant and cannot be placed here.

GOMPHIDIUS VISCIDUS Fr.—European specimens are well represented in American herbaria. The cystidia are abundant but

perhaps less so than in the American form "*columbiana*." (See remarks under the latter.)

GOMPHIDIUS spp.—One finds occasionally a few specimens rather small and slender, which are hard to place. Whether some of these are undescribed or are abnormal cannot be determined with the material and notes on hand. Two such forms are mentioned in Agaricaceae of Michigan I, p. 171–2. I have others from the Adirondack Mountains, and from North Carolina. The rarity of these, if they are autonomous species, and the chances of finding them when developed under favorable weather conditions or when not too old makes it difficult to "get a line on them."

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GOMPHIDIUS GLUTINOSUS

GOMPHIDIUS SUBROSEUS

GOMPHIDIUS TOMENTOSUS

NOTES AND BRIEF ARTICLES

Professor E. M. Gilbert, of the University of Wisconsin, visited the New York Botanical Garden in February, devoting some time to the study of the collections of the higher fungi.

Professor H. H. Whetzel, of Cornell University, spent several days during February at the New York Botanical Garden looking over the collections of Porto Rican fungi in collaboration with the latter institution on the survey of the fungous flora of Porto Rico.

In "Studies in Entomogenous Fungi. V. *Myriangium*" (Trans. Brit. Mycol. Soc. **10**: 75) T. Petch makes the following comment on *Myriangium tuberculans* Miles (Mycologia **14**: 80): "Miles has recently described *M. tuberculans*, found on *Carya illinoensis*, at Ocean Springs, Mississippi. From specimens with which he has kindly furnished me, this is *M. Curtisii*. The apothecia in the larger specimens are convex, so that the surface is facetted. The tissue is more compact than in the old herbarium specimens, and the stroma consequently less friable, again approaching *M. Montagnei*. The spores are 18–28 x 10–12 μ, rather larger than in the other available specimens, in which they do not exceed 24 x 9 μ. The specimens are accompanied by *Microcera coccophila*, the synnemata of the latter arising at the side of, or beneath, the *Myriangium*."

Collecting around St. Augustine, Florida

When the chilly winds began to blow, about the first of December, and the Virginia woods no longer yielded their harvest of mushrooms, my memory turned back to quaint old St. Augustine, nestling among the live-oaks and palmettos on the seacoast forty miles or so southeast of Jacksonville. The journey from Lynchburg required only twenty-five hours, the weather meanwhile becoming notably milder, although the hotel rooms were heated upon my arrival.

Among the fungi, the wood-destroying kinds were more in evidence, since the season for most of the fleshy kinds was practically over. A big red *Flammula*, however, was growing fresh from decayed oak roots, almost equaling some pilei of *Ganoderma Curtisii* in brilliancy. *Geaster* was entirely dried out; but a common species of hard-skinned puffball (*Scleroderma*) occurred in some abundance in all stages, showing an unusually long stem because growing in sand.

On December 6, I drove northward along the west side of the Matanzas River and collected in the groves of live-oak, interspersed with a few magnolia and hickory trees. Here was Spanish moss in abundance and the pretty little resurrection-fern (*Polypodium incanum*) growing in tufts on the oak trunks. Tree-destroying fungi were common, notably *Inonotus hirsutus*, *Ganoderma Curtisii*, *Hapalopilus gilvus* and *licnoides*, *Gloeoporus conchoides*, and several species of *Coriolus* and *Stereum*. *Armillaria mellea* had decayed, but species of *Lactaria*, *Russula*, and *Cortinarius* were perfectly new and fresh. I even found one specimen of the dark form of *Venenarius phalloides*.

Turning westward across the San Sebastian River, the live-oaks were left behind and a region of real pineland encountered, with long-leaf pines, palmettos, fan-palms, bracken fern, asters, goldenrod, etc. Here I found the same large *Flammula* seen at North Beach, several specimens of *Russula* different from those growing under the oaks, and beautiful fresh hymenophores of *Tricholoma rutilans*.

I mentioned above that a single specimen of the deadly *Amanita* was found in an oak grove. A few days later, on Anastasia Island, I found quantities of the same species colored nearly white with shades of cream and green on the disk, corresponding exactly to the last crop of the season found at Lynchburg, Virginia, in oak woods. A little distance away, several clusters of a brilliant species of *Flammula* attracted my attention. It is a large southern species partial to live-oak wood. *Pyropolyporus Calkinsii*, *Cerrenella Ravenelii*, *Pycnoporus sanguineus*, and *Pogonomyces hydnoides* were also found in the same locality under oaks and *Gloeophyllum Berkeleyi* on pine wood.

Near the great alligator farm on this island, I discovered

about two dozen clusters of the phosphorescent and poisonous species, *Clitocybe illudens*, growing from live-oak roots and stumps. Such was the brilliancy of these clusters that they suggested huge orange flowers blossoming without stalks directly from the earth.

The above brief sketch may give some idea of collecting about St. Augustine early in December. The climate here is one of the finest in the world and the whole region very charming.

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